

Four Laboratory-Associated Cases of Infection with *Escherichia coli* O157:H7

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An investigation of four cases of infection with *Escherichia coli* O157:H7 among laboratorians from different clinical laboratories revealed that the DNA fingerprint pattern of each case isolate was indistinguishable from that of an isolate handled in the laboratory prior to illness. These data suggest that the infections were laboratory acquired, and they demonstrate the importance of laboratorians strictly adhering to biosafety practices recommended for the handling of infectious materials.

Escherichia coli O157:H7 was first recognized as a human enteric pathogen in 1982 (7). Transmission occurs mainly through the ingestion of undercooked beef, raw milk, or contaminated water. Although the incidence of *E. coli* O157:H7 remains low compared to that of other enteric pathogens (6), the severity of symptoms can be pronounced, with approximately 5% of cases developing hemolytic-uremic syndrome. Four separate clinical laboratory-associated *E. coli* O157:H7 infection cases were identified in New York State (NYS) over a 5-year period from 1999 to 2004. All four individuals became ill after having manipulated *E. coli* O157:H7 isolates in their respective laboratories. Each of the four was extensively interviewed, and pulsed-field gel electrophoresis (PFGE) was conducted on the *E. coli* O157:H7 case isolates and on the corresponding laboratory culture isolates.

Isolates from the four individuals and the isolate that each person handled before onset of illness were submitted to the Wadsworth Center Bacteriology Laboratories for identification, serotyping, and PFGE testing. The isolates were identified as *Escherichia coli* based on the phenotypic characteristics, using conventional biochemicals. The isolates were serotyped as *E. coli* O157:H7 by a commercial O157 slide latex agglutination test (Oxoid Inc., Basingstoke, United Kingdom) and by H7 antisera (Becton, Dickinson and Company, Sparks, MD). PFGE was performed to determine the DNA fingerprint profile pattern. The DNA was digested with restriction endonucleases XbaI and AvrII (New England Biolabs, Beverly, MA), and the fragments were separated in 1.0% agarose gels on a homogeneous electric field apparatus (CHEF Mapper; Bio-Rad Laboratories, Richmond, CA). This assay was performed according to a standardized protocol provided by the Centers for Disease Control and Prevention (4). The DNA fingerprint profile patterns were considered to be indistinguishable, a PFGE pattern match, if all bands greater than 20 kb in size were identical.

Case 1, laboratory 1. In September 1999, a 43-year-old female laboratorian experienced symptoms of bloody diarrhea,

fever, and abdominal cramps 3 to 4 days after initial exposure in her laboratory to a stool sample positive for *E. coli* O157:H7. Case 1 had worked during the holiday weekend of a county fair *E. coli* O157:H7 outbreak, when the volume of specimens at area laboratories increased substantially (3). She wore gloves intermittently throughout the day, and her laboratory coat remained open. Case 1 was the laboratorian responsible for the identification of *E. coli* O157:H7 isolates; this included handling of agar plates, use of an automated identification system, and performance of slide latex agglutination. Vortexing of *E. coli* O157:H7 suspensions was performed on the laboratory bench top without latex gloves being worn. The laboratory telephone was answered both with and without latex gloves being worn. Hands were not washed each time that the gloves were removed, although they were always washed prior to exit from the laboratory. Case 1 did not attend the county fair, nor did any close contacts. She recalled having eaten an undercooked hamburger at a picnic prior to illness, but no other attendees of that picnic became ill. The PFGE pattern of the *E. coli* O157:H7 isolate from case 1 was indistinguishable from that of a fair attendee's isolate that had been cultured in laboratory 1 prior to the onset of her symptoms.

Case 2, laboratory 2. In July 2000, a 51-year-old female laboratorian experienced symptoms of bloody diarrhea, fever, abdominal cramps, and vomiting, 4 days after working with an *E. coli* O157:H7 isolate. Case 2 always wore latex gloves and a buttoned laboratory coat when working with isolates, changed gloves frequently, and always washed her hands after each glove removal. Case 2, wearing gloves, made a suspension of *E. coli* O157:H7 with a swab and vortexed the tube on the bench top. Gloves were removed prior to computer and telephone use. During this time period, students were undergoing training in the laboratory. Case 2 did not recall any possible exposures outside of the laboratory. The PFGE pattern of the *E. coli* O157:H7 isolate from case 2 matched that of the isolate tested in the laboratory prior to the onset of her symptoms.

Case 3, laboratory 3. In February 2003, a 26-year-old female laboratory worker experienced cramping, abdominal pain, and bloody stool, 4 days after a NYS proficiency sample containing *E. coli* O157:H7 had been received in laboratory 3. Case 3 did not wear latex gloves, but she always wore a buttoned laboratory coat. She handled the *E. coli* O157:H7 isolate to make a

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subculture only. Vortexing was not performed. Two coworkers manipulated the same isolate throughout this time period, one of whom wore latex gloves while using the computer and while turning faucet handles of a laboratory sink. These individuals did not routinely wash their hands after the removal of their latex gloves. Case 3 had eaten a cooked hamburger 6 days prior to illness, although no one else at that meal became ill. The PFGE patterns of the *E. coli* O157:H7 isolates from case 3 and the proficiency sample matched.

Case 4, laboratory 4. In March 2004, a 45-year-old female presented with diarrhea 3 days after handling an *E. coli* O157:H7 isolate. Case 4 always wore latex gloves and a buttoned laboratory coat. This laboratory worker performed a latex agglutination test to identify the *E. coli* O157:H7 isolate. Vortexing was not performed. She wore latex gloves while answering the telephone and using the computer. Her hands were washed each time that the gloves were removed. This laboratory worker had eaten cooked hamburger 1 to 2 days prior to symptoms, although no one else at the meal became ill. The PFGE patterns of the *E. coli* O157:H7 isolates from case 4 and the laboratory specimen that she had worked with were indistinguishable.

These four NYS cases are the first laboratory-associated *E. coli* O157:H7 infection cases to be published in the United States. Laboratory-associated cases have been documented in Switzerland, Scotland, and elsewhere in the United Kingdom (1, 2, 10). No specific laboratory accidents or lapses in procedure were identified in these investigations.

A review of the Wadsworth Center Bacteriology Laboratories' *E. coli* O157:H7 PFGE database of 452 isolates revealed extensive DNA profile pattern diversity. PFGE is performed and analyzed for all *E. coli* O157:H7 isolates submitted and compared to all previous strains in the database. The PFGE pattern from case 1 matched the fair outbreak pattern, which to date has been observed in NYS only during the fair outbreak. The *E. coli* O157:H7 PFGE patterns from cases 2, 3, and 4 were identical. This pattern was observed in 16 isolates from 11 counties over a 4-year period. No clusters were detected for cases 2, 3, and 4. Since this pattern is identified in approximately 4% of NYS isolates, there could be an alternative, although unlikely, source exposure from outside the workplace.

The low infectious dose of *E. coli* O157:H7 and its prolonged survival on stainless steel surfaces may have contributed to laboratory transmission in these cases (2, 8, 9). Standard laboratory biosafety practices recommended by the Centers for Disease Control and Prevention and the National Institutes of Health should be strictly adhered to at all times when potentially infectious clinical materials and cultures are handled (5).

These guidelines recommend that latex gloves be worn when hands may come in contact with potentially infectious materials. If gloves are worn during a laboratory procedure and then not appropriately removed, substantial risk exists for cross-contamination of surfaces and items. Gloves should be discarded after the procedure is completed, or if they become contaminated during the procedure. Hands should be washed thoroughly after each removal of gloves. Phones and computers should be used only after latex gloves have been removed and hands have been washed. In addition, procedures with aerosol or high splash potential, such as the vortexing of suspensions of infectious organisms, should be conducted in a biological safety cabinet.

Upon interview, the four individuals could not recall any obvious breaches in laboratory procedure prior to onset of symptoms. They did not handle stool specimens or reuse gloves. However, all four laboratorians did not strictly follow the recommended standard laboratory biosafety practices. It is the responsibility of each clinical laboratory to adhere to standard biosafety practices and guidelines, to ensure that personnel are fully trained, and to closely monitor adherence to these biosafety procedures. Strict adherence by laboratory workers to the standard laboratory biosafety recommendations will minimize the transmission of any infectious organism, including *E. coli* O157:H7, to themselves and to their coworkers.

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